

Protocol for colony PCR

Taq master mix 5ul
Forward primer 1ul
Reverse primer 1ul
dd water 2.5ul

After preparation of the reaction mix, pick a single colony and dip in the mix. The reaction condition settings are as follows:

94°C 30s
55°C 30s
72°C 1kb/min
Cycles: 30-35

Protocol for PCR with PrimeStar HS DNA polymerase

5XPrimeSTAR® Buffer 10ul
dNTP Mixture(2.5mM) 4ul
Forward primer 1ul
Reverse primer 1ul
Template <200ng
PrimeSTAR® HS (2.5 u/ul) 0.5ul
ddwater up to 50ul

In 50ul reaction system it takes much time for the sample to change from one temperature setting to another temperature setting, which has negative effect to the PCR. We suggest mix the agents listed above together then divide them into five 10ul reaction system. This procedure gives better result according to our experience.